

gradient involves the caudal third of the body in *Dero limosa*, the caudal half or more in *Lumbriculus inconstans*, and all but the first 5-15 somites in the *Tubificidæ*. Only the typical number of head somites are regenerated following amputation of varying numbers of anterior somites. The head regenerates a tail only when accompanied by a certain number of trunk somites and the tail regenerates a head only when it is of a certain minimum size whereby the gradient is eliminated. Under these conditions, regardless of length, a piece of *Dero limosa* regenerates a normal worm. If of appropriate length, any part of the body of *Lumbriculus inconstans* regenerates a normal worm, normal posterior regeneration occurring at any level. Formation of the head in *Tubifex* ceases at about the level of the fifteenth somite, while in *Limnodrilus* it ceases at the level of the seventh somite. The gradient of an axial series of pieces is not the same as that of the whole worm since temporary stimulation results from the cutting. Rate of metabolism is an important factor in anterior regeneration in short pieces since head formation will be inhibited in proportion to the metabolic rate of the old piece. If this rate be low, no inhibition of the new tissue occurs and a normal head is produced. Normal heads are always formed on long pieces since the dynamic factors are not important and the primary gradient determines the increased independence of cells at an anterior level over those of a more posterior level.

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NOTES ON THE COLLECTION AND REARING OF VOLVOX

Since the rearing of *Volvox* for laboratory use seems to be more or less unusual the writer has deemed it worth while to present here the results of some work done by a class of students and himself along this line.

Colonies of *Volvox aureus* Ehrenberg were found last fall in some abundance in small sphagnum pools on the west shore of a small glacial lake west of Ann Arbor known as the First Sister Lake. Collections were made on October 27, 1915. Small water-filled depressions in the sphagnum 10 to 30 inches in diameter were

sought out and into these depressions pint jars were thrust in such a way as to secure some bits of sphagnum and decaying vegetable matter together with water. The contents of the jars were sedimented and the liquid just above the sediment examined for colonies. When found the water at the top was poured off since the organisms were settling to the bottom and the remaining water and sediment from several jars poured together. Examination of many small pools on the shore of this lake showed that *Volvox* was not present in many of them. Some pools yielded *Pleodorina*. No colonies were found in the water of the lake itself.

Smith (1907) who has collected *Volvox* in the vicinity of Ann Arbor has found it in "small glacial pools containing *Riccia* and duckweed" but apparently had not found it among sphagnum. He (Smith, 1905) found it best in permanent pools but apparently never in great abundance. He collected it by "dipping it up together with a little of the water" and also "by sweeping a bolting cloth net over water plants, or better, using a 'Birge net'". Smith's method of dipping up the water together with plant material has been found by me to be the best method for collecting the organism here in Michigan. While in Nebraska the writer found *V. perglobator* Powers in such great abundance and in such clean open water that it was best collected with a small dipnet covered with bolting cloth or India linen. In this way large quantities of water could be passed through the net in a short time.

The collections made on October 27, 1915, were brought to the laboratory in the pint jars filled almost to the top with the water in which the organisms grew, together with about a half inch of the decaying vegetable material from the same source. Arrived at the laboratory these jars, still covered, were placed on the outside window ledge on the north side of the laboratory where no direct sunlight could strike them. They were kept here until November 29 when ice began to form on the surface of the cultures. At this time all jars except three belonging to the writer were brought inside the building. The writer's cultures remained outside for some time longer. While outside the window the number of colonies had increased greatly but when brought inside the numbers steadily diminished and soon disappeared. One student, Miss Rose Mayer,

placed her cultures in an unheated room, not in direct sunlight, where they continued to thrive for some time. An abstract from her report on her cultures is here given:

1915.

- Oct. 27. Volvox collected. (Other data as given above.)
- Oct. 28. Jars containing Volvox were placed outside of the window on the north side of the building.
- Nov. 3. Volvox was multiplying.
- Nov. 15. Still increasing in number.
- Nov. 29. Cold outdoors, some ice on surface of water. As many colonies as could be gathered were preserved. The remainder were placed in an unheated room but not in direct sunlight, i. e., the windows (south) were covered with gauze to diffuse the light.
- Dec. 13. A considerable increase had taken place. Another lot was removed and fixed. Remainder was returned to the cold room.
- Dec. 18. Colonies were quite numerous, but were beginning to fall to the bottom of the jar.
- Jan. 25. No colonies were observed. Since the last examination the temperature of the room had increased considerably.

When the cultures belonging to seven other students were brought inside they were so placed that they received some morning light. These cultures soon ran out and the writer has no further record of them. The writer's cultures (three pint jars) remained on the window ledge on the north side of the building until about December 5, but were protected at night with a cloth thrown over them. When brought inside they were placed on the window sill near the radiator. They received only north light. The colonies gradually disappeared but no record was kept of their last appearance. At various times the cultures were examined ocularly or with a hand lens but no colonies were seen until February 19 when a few colonies were seen in one jar only. They were noted again on February 26, and on March 7 the record states that they were in fair abundance. On March 16 the estimated number was 200 to 400 colonies and on March 27 several thousand were seen. At the time of writing, April 27, the water is green with them and they lie close together on the surface of the decaying vegetation.

During the fall and winter no colonies in sexual stages were seen. On March 25 a single zygote was found in the debris from the bottom of the culture. April 25 several colonies with ripe zygotes were found but they were not numerous.

Of the other two cultures one was destroyed by accident about the middle of March but no colonies had appeared in it at that time. The other culture was observed from time to time and in it 3 colonies were found March 20, about 20 on March 25, and about 50 on March 27. April 20 there were several thousand colonies present. These cultures are being kept in the hopes that further information may be gained in regard to the culture of this organism.

In conclusion it should be stated that since these cultures were not kept under controlled conditions it is possible that this success could not be repeated. However, certain points in the culture of this organism may be emphasized, viz., the water for the cultures should be from the same source as the organisms. Never use tap-water for making up the culture or for making good evaporation. Keep the cultures covered to prevent evaporation and consequent change in density of the medium and to exclude the dust and bacteria. The presence of organic material seems to be beneficial. Direct sunlight is unnecessary and is to be avoided because it causes too great variations in temperature in closed vessels. North light is good; in fact many algæ thrive in it as evidenced by the good growth of algæ in these cultures. Low temperature, above freezing, in early winter seemed to favor development. Old cultures should not be destroyed unless they have become hopelessly foul but they should be kept and the organisms given a chance to reappear.

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A NEW EMBEDDING STAGE

A new electrically heated embedding stage prepared according to designs prepared by laboratory men in this university has been recently put on the market by Eberbach & Co. of Ann Arbor. The essential parts of this embedding stage (see cut, Pl. XXI, Fig. 2) are a transite base $17\frac{3}{4}$ inches long by $4\frac{1}{2}$ inches wide mounted on three levelling screws, a copper stage made in two parts, 4 by 13 inches and 4 by 4 inches respectively, and under one end of the longer copper stage an electric heating unit. The heating unit may be wound for any voltage and to yield any desired temperature. Those in use in the Zoölogical Laboratory are designed for 110 v. alternating or direct current and the current requirement is 0.5 ampere. This yields a temperature of about 74° C. No regulator or rheostat or other provision for controlling or varying the temperature is provided but since the coil is situated under one end of the stage lower temperatures may be secured by moving the object away from coil. A scale to indicate the gradations of temperature could be attached if desired. In practice the coil is attached to a convenient electric receptacle near the paraffine bath and that part of the stage over the coil is heated sufficiently to melt paraffine in a few minutes. The embedding tray may now be warmed over the hot stage, filled with melted paraffine and moved to a point on the stage where the paraffine is kept just melted. Objects to be embedded are now transferred to the embedding tray, oriented, and the label inserted at the end of the tray with the legend towards the margin of the tray. Now the tray is gently moved to the unheated end of the stage where the paraffine is permitted to congeal on the bottom sufficiently to hold the objects in place. Then the tray is trans-